Fecal microbiota transplant: A novel biological approach to extensively drug-resistant organism-related non-relapse mortality.

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Summary

Extensively drug-resistant organisms (XDRO) are a global threat to health. Colonisation with XDRO prior to hematopoietic cell transplantation (HCT) frequently results in delayed delivery of antimicrobials to which the organisms are susceptible and significantly increases non-relapse mortality. Their inherent resistance to available antimicrobial agents coupled with a preponderance to evolve further resistance makes biological approaches attractive. Suppression of pathogenic organisms by fecal microbiome transplantation has previously been demonstrated, and here we detail use of this approach to successfully supress XDRO prior to HCT that permitted an uneventful transplant course in an otherwise high-risk situation.

Non-relapse mortality (NRM) of allogeneic hematopoietic cell transplantation (HCT) has progressively fallen over the last four decades. Better supportive care, particularly in managing infection has significantly contributed to the improved safety over that period. However, antimicrobial resistance poses a significant global threat to health (1), and the emergence of extensively drug-resistant organisms (XDRO) within HCT units now poses a direct threat to transplant recipients (2). Gut colonisation with XDRO has been associated with an inased NRM (3) and infections with XDRO during neutropenic periods are complex to manage and associated with a high mortality (2). Innovative approaches in preventing and managing them are therefore necessary to avoid reversing much of the progress made in limiting NRM over the last 4 decades.

A 63-year-old man presented to our institution with a new diagnosis of Philadelphia positive acute lymphoblastic leukemia and received treatment following the UKALLXII trial schedule (4). He achieved complete remission after induction chemotherapy together with imatinib. Following intensification chemotherapy and continuous imatinib, allogeneic HCT was recommended to consolidate his therapy. His treatment course was complicated by two separate episodes of extended-spectrum beta-lactamase (ESBL)-producing *Escherichia coli* bloodstream infections, two episodes of *Clostridium difficile* infection (CDI), and central line-related methicillin sensitive *Staphylococcus aureus* bacteremia. Each infection was successfully treated with antimicrobials, but he was subsequently found to be colonised with a highly-resistant *ges-5* carbapenemase-producing Enterobacteriaceae (CPE), *Klebsiella oxytoca,* on routine rectal screening (table 1).

While gut colonisation with XDRO does not pose any significant risk *per se*, these organisms can cause opportunistic infection during periods of prolonged neutropenia. Rates of spontaneous clearance of these organism from colonised individuals are low, even in immunocompetent hosts, ranging from 7-30% (5,6). Treatment options for elimination of XDRO from their site of origin within the intestine are limited; non-absorbable antimicrobial agents often lead to only transient suppression (5), and may precipitate the development of further resistance. Given the success of donor fecal microbiota transplant (FMT) in the management of recurrent/refractory CDI (7), and the apparently acceptable safety profile when used for CDI in the HCT setting (9), there is considerable interest in the potential role of FMT in gut decontamination prior to HCT. Recipients of FMT for CDI have been shown to have fewer antibiotic-resistant organisms within their gut microbiota following transplantation (10) and there are emerging clinical reports of successful use of FMT in gut decontamination of a variety of XDRO (including ESBL and CPE) (11), even in the setting of haematological disorders (8). Therefore after discussion, this patient was offered FMT prior to

allogeneic HCT in an attempt to eradicate the XDRO and *C. difficile* from its intestinal niche, with the aim of minimising his HCT NRM.

Following informed consent, the patient received gut preparation with four days of oral vancomycin and neomycin, both 500mg four times daily. Antibiotics were stopped 24 hours prior to FMT delivery, and preparation was completed with iso-osmotic bowel purgatives (Kleen Prep). The unrelated donor stool was pre-screened, and negative for *C. difficile* PCR and toxin, as well as for XDRO; other routine donor screening for transmissible infection was also negative (12). Preparation of the transplant occurred immediately after stool donation under strict anaerobic conditions, using an adapted version of a previously-described protocol (13) and stored at -80°C until required. The FMT product comprised a thawed slurry of around 100ml homogenised stool preserved in a mixture of glycerol and phosphate buffered saline (15:85, v/v) and was delivered via nasogastric tube. Fasting was instituted six hours prior to receipt of the FMT, and treatment with a proton-pump inhibitor (omeprazole) and pro-kinetic (metoclopramide) were administered one hour prior to FMT delivery. The patient was allowed to eat and drink normally one-hour post-administration. Following the procedure, he experienced mild nausea, loose stool and abdominal discomfort, which all resolved after 24 hours without any specific intervention. Repeat rectal screening 7 days following the FMT showed continued carriage of the ESBL *E. coli* but no evidence of *ges-5 K. oxytoca* CPE or *C. difficile*. By day 16 after FMT neither the CPE nor ESBL were detected on rectal screening swabs (Table 1).

Two weeks after FMT, the patient underwent a fludarabine (30mg/m^2 D-7 to -3) and melphalan (140mg/m2 day -2) conditioned reduced intensity sibling allogeneic HCT, with standard cyclosporine and methotrexate graft-versus-host disease (GvHD) prophylaxis. The transplant course was complicated by one episode of neutropenic fevers on day +5, with isolation of a fully-sensitive *Enterococcus faecalis* from blood cultures (table 1). Empirical treatment with piperacillin-tazobactam (4.5g three times daily), amikacin (15mg/kg once daily), teicoplanin (12mg/kg twice daily for three doses, followed by 12mg/kg once daily) as per local policy with addition of colistin (3 million units twice daily) resulted in prompt resolution of fever within 24 hours, and following isolation of the sensitive organism antimicrobials were de-escalated to piperacillin-tazobactram and teicoplanin. A second episode of neutropenic fever developed on day +10, and responded to a change in antimicrobials from piperacillin-tazobactam to meropenem (1g three times daily), and cultures remained sterile. Neutrophil engraftment was achieved on day +25 and the patient was discharged from hospital on day +29. At day +100 he was well, with no evidence of leukemia, GvHD or XDRO by rectal screening. At 12-months post-transplant the patient remains well and in remission.

Carbapenemase-producing micro-organisms are now endemic in a number of countries (1,14) and the preponderance of these organism to extend their resistance spectrums is now contributing to the emergence of strains resistance to our last resorts antimicrobials (15). A paucity in novel antimicrobials means that current approaches are restricted to minimising the risk of XDRO colonisation by antimicrobial stewardship and infection control, as well as managing clinical infection with complex, and often more toxic, antimicrobial schedules. Novel strategies are therefore required, and biological approaches would seem most favourable given the weaknesses of our current pharmacological armoury. Resident gut commensals are adapted to the intestinal microenvironment and have developed complex ecological networks upon which they have subsequently become interdependent. Pathogens are equally reliant on their microenvironment, and competition for critical nutrients, alteration of pH or oxygen tension, and production of toxic metabolites are all mechanisms by which healthy commensals are capable of supressing pathogens (16). While FMT has been reported in decontamination of XDRO in immunocompromised (17) patients and those with blood disorders before (8) here we detail our use of this biological approach in the suppression of XDROs in order to minimise NRM prior to allogeneic HCT. Our experience supports the use of FMT in this setting as safe and tolerable, and warrants further study of efficacy in a randomised fashion. The suppression of XDRO by FMT pre-HCT is particularly pertinent because rather than simply identifying an addition risk factor for NRM, the presence of XDROs should been considered a potentially modifiable risk factor, and this distinction is exceptionally important in risk stratification.

Legend

Table 1. Microbiological sample results/Timeline. *E.Coli*, *Escherichia coli*, *K. Oxytoca, Klebsiella Oxytoca*, *S. aureus, staphylococcus aureus*, *E. Faecalis, Enterococcus faecalis*, R, resistant, S, susceptible, I, intermediate, C. difficle, Clostridium difficle, PCR, Polymerase chain reaction, HCT, hematopoietic cell transplantation, XRDO, extensively drug-resistant organism.

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Days post FMT	-224	-209	-203	-177	-168	-164	-30	-30	-30	-	0	14	16	16	19	23	29	36
Sample source	Blood cultures x 2	Stool	Blood cultures x 2	Stool	Rectal screen	Rectal screen	Blood cultures & line tip	Rectal screen x 2	Rectal screen x 2	Faecal microbiota transplant	Rectal screen	Reduced intensity sibling HCT	Rectal screen	Stool	Blood cultures	Rectal screen	Rectal screen	Rectal screen
Organism	*E. coli*	*C. difficile* PCR positive	*E. coli*	*C. difficile* PCR positive	*K.oxytoca GES-5*	*K.oxytoca GES-5*	*S. aureus*	*E. coli*	*E. coli*		*E. coli*	No XDRO identified	No XDRO identified	*C. difficile* PCR negative	*E. faecalis*	No XDRO identified	No XDRO identified	No XDRO identified
Amikacin	S		S		S	S	-	S	S		S				-			
Amoxicillin	R		R		R	R	-	R	R		R				S			
Aztreonam	R		R		R	R	-	R	R		R				-			
Cefoxitin	R		R		R	R	-	R	R		R				-			
Ceftazidime	R		R		R	R	-	R	R		R				-			
Ceftriazone	R		R		R	R	-	R	R		R				-			
Cefuroxime	R		R		R	R	-	R	R		R				-			
Ciprofloxacin	R		R		R	R	S	R	R		R				-			
Co-Amoxiclav	R		R		R	R	-	R	R		R				-			
Collistin	S		S		S	S	-	S	S		S				-			
Ertapenem	S		S		R	R	-	S	S		S				-			
Gentamicin	R		R		R	R	S	R	R		R				-			
Meropenem	S		S		I	I	-	S	S		S				-			
Piperacillin-tazobactam	I		I		R	R	-	R	R		R				-			
Temocillin	R		R		R	R	-	R	R		R				-			
Tigecycline	S		S		S	S	-	S	S		S				-			
Tobramycin	R		R		R	R	-	R	R		R				-			
Trimethoprim	R		R		R	R	S	R	R		R				-			
Clindamycin	-		-		-	-	S	-	-		-				-			
Erythromycin	-		-		-	-	S	-	-		-				-			
Flucloxacillin	-		-		-	-	S	-	-		-				-			
Fusidic acid	-		-		-	-	S	-	-		-				-			
Oxacillin	-		-		-	-	S	-	-		-				-			
Penicillin	-		-		-	-	R	-	-		-				-			
Rifampicin	-		-		-	-	S	-	-		-				-			
Teicoplainin	-		-		-	-	S	-	-		-				S			
Tetracycline	-		-		-	-	S	-	-		-				-			
Vancomycin	-		-		-	-	S	-	-		-				S			